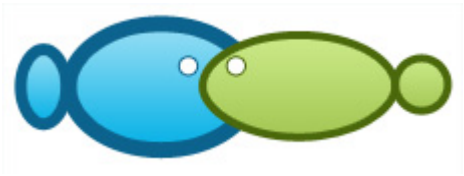

Effect of salinity on the survival of mangrove crab *Scylla tranquebarica* larvae at zoea-megalopa stages

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Abstract. The main concern in mangrove crab hatcheries is the low survival of larvae, particularly on the zoea and megalopa stages. This study aimed to determine the optimum salinity in the maintenance of mangrove crab larvae, *Scylla tranquebarica*. This study was conducted at Hall of Brackish Water Aquaculture, South Sulawesi, Indonesia. The experimental animal used was *S. tranquebarica* larvae at mite zoea-1 stage. The research container used was a black plastic basin with a capacity of 40 L filled with water media of 30 L. The feed used was natural food in the form of rotifer and nauplius *artemia*. The study was designed using a complete randomized design consisting of 4 salinity treatments with each of 3 replications namely 26, 29, 32, and 35 ppt. The data obtained were analyzed by using variance analysis and followed by further test of W-Tuckey. The results showed that salinity had a very significant effect ($p<0.01$) on osmotic rate and survival of mangrove crab larvae. The salinity of 29 ppt media resulted in osmotic rate of 54.09 mOsm/L H_2O with the highest survival of *S. tranquebarica* larvae of 33.73%. The optimum salinity for maintenance of *S. tranquebarica* larvae was in the range 30.48-30.89 ppt.

Key Words: crab cultivation, hatchery, osmotic, survival, *S. tranquebarica*.

Introduction. *Scylla tranquebarica* is one of four species of mangrove crabs that are commonly found in Indonesian waters, especially in mangrove areas. According to Pedapoli & Ramudu (2014), *S. tranquebarica* has comparative advantages such as the growth which may reach weight of 1.5 kg, disease resistance, good market demand, wide tolerance for temperature and salinity, and greater scope in export markets and the domestic market. Therefore, this type of crab is potential to be developed. Recent crab cultivation studies have been conducted on male monosex crabs in mangrove areas (Karim et al 2016), mRNA observations in mandibular organs (Tahya et al 2016a; Sunarti et al 2016), and the role of mandibular organs in supporting cultivation activities (Tahya et al 2016b; Tahya 2016).

One of the determinants of successful crab cultivation is the availability of seeds. During this time, the need of crab seeds is mostly still met from the results of arrests in nature that are fluctuating. In order to meet the needs of crab seeds, efforts have been made to produce seeds through hatcheries. Hatching of *S. tranquebarica* has been performed in several hatcheries; however, the main constraint faced is still low survival of larvae, particularly in zoea and megalopa stages. Several studies obtained a low survival of crab larvae, among others Quinitio et al (2001) obtained survival of crab larvae from zoea to megalopa stages of 0.15 to 0.74%, Jantrarotai et al (2002) obtained 7.08-22.91%, Nghia et al (2007) obtained 8-13%, Thirunavukkarasu et al (2014) obtained 12% crab larvae reaching megalopa stage.

The low survival of crab larvae is due to an inadequate maintenance environment. Therefore, to improve the larval survival, it is necessary to improve the management of seeding. Such management improvements can be made by optimizing the hatchery maintenance. One of the environmental factors that affect the maintenance of mangrove

crab larvae is salinity. According to Pedapoli & Ramudu (2014), salinity is one of the water quality parameters that affect the survival, feed efficiency, growth and health of crabs. Several studies on the salinity of crab larvae have been conducted which reported that salinity is very influential on the survival of crab larval (Pedapoli & Ramudu 2014; Gunarto & Perenrengi 2014; Mia & Shah 2010).

Adult mangrove crabs belong to euryhaline aquatic organisms with a salinity range of 1-42 ppt (Karim 2013), however, the optimum salinity range was narrower for larval size, particularly in zoea stadia (Nguyen & Truong 2004; Karim 2013). Under unoptimal salinity conditions, it can cause osmotic stress such as increased oxygen consumption, ammonia excretion, and high use of energy (Romano & Zeng 2006).

Because salinity strongly affects larval life, therefore, this study aimed to determine the optimum salinity in the maintenance of *S. tranquebarica* larvae.

Material and Method. The research was conducted at the Hall of Brackish Water Aquaculture, South Sulawesi, Indonesia. The experimental animal was crab larvae (*S. tranquebarica*) zoea-1 stage that was maintained until megalopa stage. The container used in this study was a black plastic basin filled with water media of 30 L and aerated.

The natural feeds used were rotifer and nauplius *artemia*, which was given with a density of 20 ind/mL from the zoea-1 stage to the beginning of zoea-4 stage. Once entered zoea-3 stage, it was added with *Artemia* nauplii with a density of 2 ind/mL and increased to 4 ind/mL when entered zoea-4 to zoea-5 stages.

The study was designed using a complete randomized design consisting of 4 salinity treatments with 3 replicates (12 experimental units). The four salinity concentrations tested were 26, 29, 32, and 35 ppt. The parameters studied were osmotic rate and survival of crab larvae

The osmotic rate was calculated according to the instructions of Lignot et al (2000) as follows:

$$TKO = OM - OH$$

Where:
TKO = osmotic rate (mOsm/L H_2O);
OM = media osmolarity (mOsm/L H_2O);
OH = osmolarity of hemolympha (mOsm/L H_2O).

Survival of crab larvae was calculated using the following formula:

$$S = (N_t/N_o) \times 100$$

Where:
S = survival of crab larvae (%);
N_t = number of larvae in the end of study (tail);
No = number of larvae in the beginning of study (tail).

Data were analyzed using variance analysis and further tested using W-Tuckey.

Results and Discussion. Measurement of osmotic rate of *S. tranquebarica* larvae at various salinities is presented in Table 1.

Table 1

Osmotic rate of *Scylla tranquebarica* larvae at various salinities

Salinity (ppt)	*Osmotic rate (mOsm/L H_2O)*
26	102.94 ± 4.73^{a}
29	54.09 ± 0.27^{c}
32	69.39 ± 2.28^{b}
35	98.73 ± 6.07^{b}

Different letters on different column show significant difference among treatments 5% ($p<0.05$) level.

Salinity media is the determinant of osmotic rate of crab larvae. The osmotic properties of the medium depend on the dissolved ions in the medium. The greater amount of

dissolved ions in the media causes an increase in the osmotic pressure of the media. The osmotic rate experienced by crab larvae is proportional to the difference in osmolarity between media and hemolymph. On the media with osmotic rate outside the isoosmotic range, crab larvae perform osmotic work for osmoregulation purposes,

Salinity media is very significant ($p<0.01$) in affecting the osmotic rate of crab. The highest osmotic rate was produced on the media with salinity of 26 and 35 ppt, while lowest osmotic rate was produced on the media with salinity of 29 ppt. The relationship of salinity media and osmotic rate of crabs was obtained using quadratic pattern with regression equation Y = 2074.4000-132.3800x + 2.1717x2, R2 = 0.90. By the equation, it was predicted that osmotic rate of *S. tranquebarica* larvae was at a minimum salinity of 30.48 ppt (Figure 1).

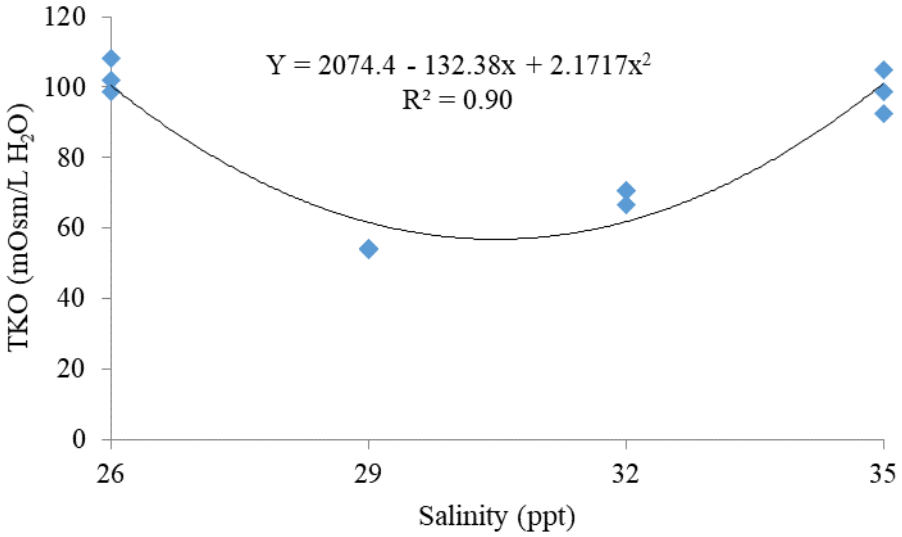

Figure 1. The relationship curve between salinity and osmotic rate of *Scylla tranquebarica*.

In the medium with salinity of 26 ppt, crab larvae are hyperosmotic to the medium. Under this condition, water from external media tends to penetrate into the thin body of the larvae. The ions tend to diffuse outward of the body and the internal fluid will be threatened with lack of ion through excretion. To overcome this problem, crab larvae will try to maintain the stability of the osmolarity of the body fluid through hyperosmotic regulatory mechanism by increasing the absorption of ions (salts) from external media through the body surface and producing hypoosmotic urine through the excretory organs (antenna glands). Furthermore, Mudagandur et al (2016) revealed that salinity causes internal body alterations.

In 35 ppt salinity medium the body fluids of crab larvae are hypoosmotic to the medium. Therefore, water from the larval body fluids tends to move out through osmosis. In this condition, crab larvae will try to maintain the osmolarity of body fluids so that the internal fluid does not move out from its cells and prevent urine fluids are not more concentrated than its hemolymph. For this purpose, crab larvae will extract H_20 from the medium by drinking or entering water through the gills and body surface. In crustaceans, the most permeable surface on the gill is a major part which is involved in osmoregulation process (Mudagandur et al 2016), whereas Freire et al (2011) revealed that high hepatopancreas and muscle activity occur in crabs which maintained at high salinity.

Survival of *S. tranquebarica* larvae at various salinities is presented in Table 2.

Table 2

Survival of *Scylla tranquebarica* larvae at various salinities

Salinity (ppt)	*Survival (%)*
26	12.16±0.97[d]
29	33.73±0.70[a]
32	29.96±1.98[b]
35	19.29±0.68[c]

Different letters on different column show significant difference among treatments 5% ($p<0.05$) level.

Salinity of the media had a very significant effect ($p<0.01$) on the survival of crab *S. tranquebarica* larvae. The highest survival of crab larvae was observed at salinity of 29 ppt and the lowest at 26 ppt. The highest survival was produced on the medium with salinity of 29 ppt which indicated that the medium supports the life of crab larvae optimally. This condition give an advantage for crab larvae in which the larvae have a better ability in dealing with osmotic stress so that they have more chance to live. Meanwhile, medium with salinity of 29 ppt has a low osmotic rate, thus there is sufficient energy for crab larvae to maintain its survival, whereas medium with salinity of 26 ppt had a high osmotic rate that led to the high use of energy. The survival values obtained in this study ranged from 12.16% to 33.73%. Several studies including Azam & Narayan (2013) obtained survival of *S. serrata* until megalopa stage of 9%. Dien (2010) obtained survival of *S. serrata* of 3.33-8.78%, Thirunavukkarasu et al (2014) obtained survival of *S. tranquebarica* of 12.5%, Hassan et al (2011) obtained survival of *S. serrata* of 9.5-18%.

Salinity is one of the most important abiotic factors that affect the aquatic organisms. Therefore, it is necessary to determine the optimum salinity for crab larvae to suit its need in maintaining its survival. According to Pedapoli & Ramudu (2014), the optimum survival for crab larvae is in the range of 15-25 ppt. The low survival of an organism due to salinity change is a reflection capacity loss of the organism to osmotic changes. The optimum salinity requirements of each aquatic organism vary as found in crab larvae *(S. olivacea)* ranging from 28 ppt to 30 ppt (Gunarto & Perenrengi 2014). Mia & Shah (2010) reported the best survival rate of *S. serrata* at salinity of 25 ppt.

The relationship between media salinity and survival of crab *S. tranquebarica* larvae was patterned quadratic with the equation Y = 813.620 + 54.994x-0.892x2 with R2 = 0.93. Based on the regression equation, it could be predicted that the optimum salinity resulting in maximum survival of crab larvae was at salinity of 30.89 ppt (Figure 2).

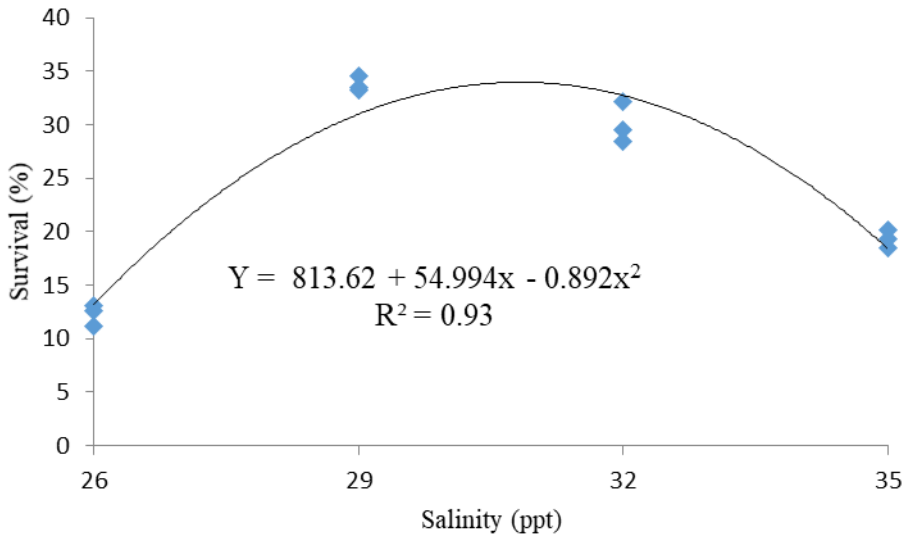

Figure 2. Relationship curve between salinity and survival of crab *Scylla tranquebarica* larvae.

Water quality. During the research, it was measured chemical and physical parameters including temperature, dissolved oxygen, pH, and ammonia concentration. The media temperature for all treatments ranged between 29-30°C, dissolved oxygen was of 4.3-4.7 ppm, pH 7.7-7.9 and ammonia concentration of 0.003-0.005 ppm. Those values are still within a reasonable range to support the life of *S. tranquebarica* larvae. According to Karim (2008) optimum salinity for the needs of mangrove crab metabolism is 25 ppt.

Conclusions. Medium with salinity of 29 ppt results in a working osmotic rate by 54.09 mOsm/L H_2O with highest survival of *S. tranquebarica* larvae by 33.73%. Optimum salinity for crab larvae maintenance is in the range of 30.48-30.89 ppt.

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